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Mechanistic Study of Inhibition of Levofloxacin Absorption by Aluminum Hydroxide

MAKOTO TANAKA,* TADASHI KURATA, CHIHO FUJISAWA, YUMI OHSHIMA, HIROYUKI AOKI, OSAMU OKAZAKI, AND HIDEO HAKUSUI

Developmental Research Laboratories, Drug Metabolism and Analytical Chemistry Research Center, Daiichi Pharmaceutical Co. Ltd., 1-16-13, Kitakasai, Edogawa-ku, Tokyo 134, Japan

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The mechanisms of reduction in absorption of levofloxacin (LVFX) by coadministration of aluminum hydroxide were studied. The partition coefficient of LVFX (0.1 mM) between chloroform and phosphate buffer (pH 5.0) was reduced by 60 to 70% with the addition of metal ions such as Cu^{2+}, Al^{3+}, and Fe^{2+} (0.8 mM), which indicated the formation of LVFX-metal ion chelates. However, there was no significant difference in absorption from rat intestine between the synthetic LVFX-Al^{3+} (1:1) chelate (6.75 mM) and LVFX (6.75 mM) in an in situ recirculation experiment. On the other hand, $Al(NO_3)_3$ (1.5 mM) significantly inhibited the absorption of LVFX (1.5 mM) by 20% of the control in the in situ ligated loop experiment, in which partial precipitation of aluminum hydroxide was observed in the dosing solution. Data for adsorption of LVFX and ofloxacin (OFLX) from aqueous solution by aluminum hydroxide were shown to fit Langmuir plots, and the adsorptive capacities (r_{max}) and the K values were 7.0 mg/g and 1.77×10^4 M^{-1} for LVFX and 7.4 mg/g and 1.42×10^4 M^{-1} for OFLX, respectively. The rate of adsorption of several quinolones (50 μM) onto aluminum hydroxide (2.5 mg/ml) followed the order norfloxacin (NFLX) (72.0%) > enoxacin (ENX) (61.0%) > OFLX (47.2%) ≈ LVFX (48.1%). The elution rate of adsorbed quinolones with water followed the rank order LVFX (17.9%) ≈ OFLX (20.9%) ≈ ENX (18.3%) > NFLX (11.9%). These results strongly suggest that adsorption of quinolones by aluminum hydroxide reprecipitated in the small intestine would play an important role in the reduced bioavailability of quinolones after coadministration with aluminum-containing antacids.

Levofloxacin (LVFX), (−)-(*S*)-9-fluoro-2,3-dihydro-3-methyl-10-(4-methyl-1-piperazinyl)-7-oxo-7*H*-pyrido[1,2,3-*de*][1,4]benzoxazine-6-carboxylic acid hemihydrate, is a new quinolone antimicrobial agent which exhibits broad-spectrum in vitro bactericidal activities against gram-positive and gram-negative aerobes (8, 29). LVFX is the more antibacterially active optical isomer of racemic ofloxacin (OFLX) (8).

The bioavailability of quinolone antimicrobial agents including LVFX has been shown to be less when they are ingested with antacids or mineral preparations (13, 24), and the binding of metal ions contained in these preparations to the 4-keto- and 3-carboxyl-groups of quinolones to form nonabsorbable chelates has been suggested as the possible mechanism responsible for the reduced absorption of quinolones (4, 10, 17, 19, 25). However, attempts to relate the magnitude of reduction in bioavailability by antacids to chemical structures of quinolones or to chelate formation constants have been generally unsuccessful, and the mechanism remains to be elucidated.

For the present paper, we studied the mechanisms of pharmacokinetic interaction of LVFX with aluminum hydroxide. The influence of metal ions on the partition coefficient of LVFX between chloroform and phosphate buffer and the adsorptive characteristics of various quinolones such as LVFX, OFLX, enoxacin (ENX), and norfloxacin (NFLX) on the surface of dried aluminum hydroxide gel were studied. The inhibitory effect of the Al^{3+} ion on LVFX absorption in rats was investigated by in situ recirculation and the ligated intestinal loop method.

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MATERIALS AND METHODS

Chemicals. OFLX, LVFX, and LVFX-Al^{3+} (1:1) chelate were synthesized in the Research Institute of Daiichi Pharmaceutical Co., Ltd. (1, 8, 22). ENX (Dainippon Pharmaceutical Co., Ltd., Osaka, Japan), NFLX (Kyorin Pharmaceutical Co., Ltd., Tokyo, Japan), and dried aluminum hydroxide gel (Chugai Pharmaceutical Co., Ltd., Tokyo, Japan) were also used in this study. [^{14}C]LVFX (specific radioactivity, 2.76 MBq/mg) was purchased from Daiichi Pure Chemicals Co., Ltd. (Tokyo, Japan). Its radiochemical purity was more than 99.3% as checked by thin-layer chromatography. *P*-[G-^{3}H]-acetaminophen (370 MBq/mg) and γ-[U-^{14}C]-γ-aminobutyric acid (GABA) (54.3 MBq/mg) were purchased from New England Nuclear (Boston, Mass.). All other reagents were of analytical grade and used without further purification.

Partition study. The salts [$CuSO_4$, $Al(NO_3)_3$, $FeSO_4$, $MgSO_4$, $ZnSO_4$, and $Ca(NO_3)_2$] and LVFX were dissolved in phosphate buffer (pH 5.0). The metal ion solution (0 to 2 mM) (1 ml) and LVFX solution (0.2 mM) (1 ml) were mixed in a glass-stoppered test tube by shaking at room temperature for 30 min. The resulting mixture was extracted with chloroform (2 ml) by shaking for 10 min and centrifuged at 1,800 × *g* for 10 min. The concentration of LVFX in the organic layer was determined by high-performance liquid chromatography (HPLC).

Absorption study with rats. (i) In situ recirculation method. Male Sprague-Dawley rats (7 weeks old) kept fasting overnight were anesthetized with sodium pentobarbital (15 mg per rat) given by intraperitoneal injection. A recirculating

* Corresponding author.

perfusion technique was used (28). A 15-cm segment of the intestine was cannulated at the duodenum and jejunum, prewashed with 5 ml of saline kept at 37°C, and perfused with the solution of LVFX or LVFX-Al^{3+} (1:1) chelate at a known initial concentration (6.75 mM), both solutions being preheated to 37°C and maintained at this temperature. The perfusing medium was phosphate buffer (pH 5.0), and the drug solution was recirculated at a rate of 5 ml/min with a peristaltic pump. Successive samples (0.2 ml) were taken at 10, 20, 30, 45, and 60 min after the start of perfusion.

(ii) In situ ligated intestinal loop method. Male Sprague-Dawley rats (7 weeks old) were used in this study. The animals were kept fasting overnight. A loop approximately 5 cm long was prepared from the upper small intestine. The loops were injected with 0.2 ml of [^{14}C]LVFX, [^{3}H]GABA, or [^{14}C]acetaminophen in phosphate buffer (pH 6.5) (1.5 mM) containing $Al(NO_3)_3$ (0, 1.5, or 6.0 mM) and replaced in the abdominal cavity for 15 min. The loops were removed, and the radioactivity remaining in the loop contents was measured by a liquid scintillation counter.

Adsorption study. The dried aluminum hydroxide gel was suspended in distilled water (5 mg/ml). LVFX and OFLX were dissolved in water by adding a small amount of phosphoric acid, if necessary. The antacid suspension (0.5 ml) and quinolone solution (50 to 1,000 μM) (0.5 ml) were mixed and incubated for 1 h in a shaking incubator with a thermostat at 37°C. At the end of the 1-h period, samples were centrifuged at 23,000 × *g* for 30 min, and the drug concentrations in supernatant were determined by HPLC. The amount of drug adsorbed by aluminum hydroxide was calculated by determining the difference between the amount initially added and the amount remaining in the supernatant. The average of the three determinations was used for data analysis.

The aqueous solutions of LVFX, OFLX, ENX, and NFLX (100 μM) (0.5 ml) were mixed with aluminum hydroxide suspension (5 mg/ml) (0.5 ml) to determine the percent binding by aluminum hydroxide of individual quinolones. The procedure was the same as that described above.

Treatment of binding data. The results obtained with LVFX and OFLX (see Fig. 3) suggested that the binding of the various quinolones to aluminum hydroxide could be analyzed by assuming that the binding is analogous to the adsorption of a drug on a solid and follows a type I Langmuir isotherm. Specifically, the amount of drug bound per milligram of aluminum hydroxide, r, should be related to the concentration of free drug, $[D]_{free}$, by

$$r = \frac{r_{max} K [D]_{free}}{1 + K [D]_{free}}$$

where r_{max} is the maximum moles bound per milligram of aluminum hydroxide, that is, the amount needed to form a monolayer, and K is a constant related to the affinity or strength of the interaction. Rearrangement of this equation to

$$\frac{1}{r} = \frac{1}{r_{max}} + \frac{1}{r_{max} K} \cdot \frac{1}{[D]_{free}}$$

shows that a linear relation should be obtained when $1/r$ is plotted as a function of $1/[D]_{free}$. The extrapolated y-axis intercept is $1/r_{max}$, and the slope is $1/r_{max}K$. Thus, such a treatment of the binding data would permit a calculation of r_{max} and K for each drug and provide a basis for comparisons of the binding of drugs to aluminum hydroxide.

Elution study. The elution of LVFX, OFLX, ENX, and NFLX from aluminum hydroxide was determined by washing the residue after the adsorption run with distilled water (1 ml). The residue resuspended in water had been shaken at 37°C for 1 h. The supernatant concentrations of quinolones eluted were determined by HPLC after centrifugation at 23,000 rpm for 30 min.

Drug assay. The concentrations of quinolones in samples were determined by HPLC, with a modification of the method of Okazaki et al. (18). The high-performance liquid chromatograph (Model 655A-11; Hitachi, Tokyo, Japan) was equipped with a variable-wavelength UV detector (Model 655A; Hitachi). The wavelengths were set at 295 nm for OFLX and LVFX and 280 nm for ENX and NFLX. Separation was achieved on a reversed-phase column (TSK-GEL ODS-80 TM, 5 μm, 150 by 4.6 mm in inside diameter; Tosoh, Tokyo, Japan) with a mobile phase consisting of tetrahydrofuran–50 mM phosphate buffer (pH 2.0)–1 M ammonium acetate (8:92:1 ratio for OFLX and LVFX and 4:96:1 ratio for ENX and NFLX) at the flow rate of 1.0 ml/min. ENX was used as the internal standard for the determination of NFLX, and NFLX was employed as the internal standard for ENX.

The organic layer obtained in the partition study was diluted with the methanol solution of the internal standard, and the resulting mixture was evaporated to dryness under reduced pressure. The residue dissolved in the mobile phase was applied to HPLC.

The perfusate in the absorption study or the supernatant in the adsorption study was diluted with aqueous solutions of internal standards, and the resulting mixture was directly injected into HPLC.

Statistical analysis. Statistical analyses were performed with Student's t test with $P = 0.05$ as the minimal level of significance.

RESULTS

LVFX partitioned between chloroform and phosphate buffer (pH 5.0) with a partition coefficient of 0.65. With increasing concentrations of various cations, the partition coefficients decreased as shown in Fig. 1. The partition coefficient of LVFX (0.1 mM) was reduced by 60 to 70% with the addition of metal ions such as Cu^{2+}, Al^{3+}, and Fe^{2+} (0.8 mM), which indicated the formation of LVFX-metal ion chelates. On the other hand, Mg^{2+}, Zn^{2+}, and Ca^{2+} (0.8 mM) showed much weaker effects and reduced the partition coefficient only by less than 22%.

The absorption of LVFX-Al^{3+} (1:1) chelate in rats was examined by the in situ recirculation method. The pH of perfusing medium was adjusted to 5.0 because of the higher stability of the chelate in the lower pH region (1). The absorption of LVFX and the chelate is shown in Table 1, which indicates that there is no significant difference in absorption from the rat intestine between LVFX and its chelate.

The effect of the Al^{3+} ion on absorption of LVFX in rats was also investigated by the in situ ligated intestinal loop method. The pH of the dosing solution was adjusted to the value found for the proximal intestine in rats or 6.5 (12). Al^{3+} (6.0 mM) showed no inhibitory effect on the absorption of acetaminophen and GABA (1.5 mM), which were model compounds for passive and active absorption, respectively (Fig. 2). On the other hand, Al^{3+} (1.5 mM) significantly inhibited the absorption of LVFX (1.5 mM) by 20% of the

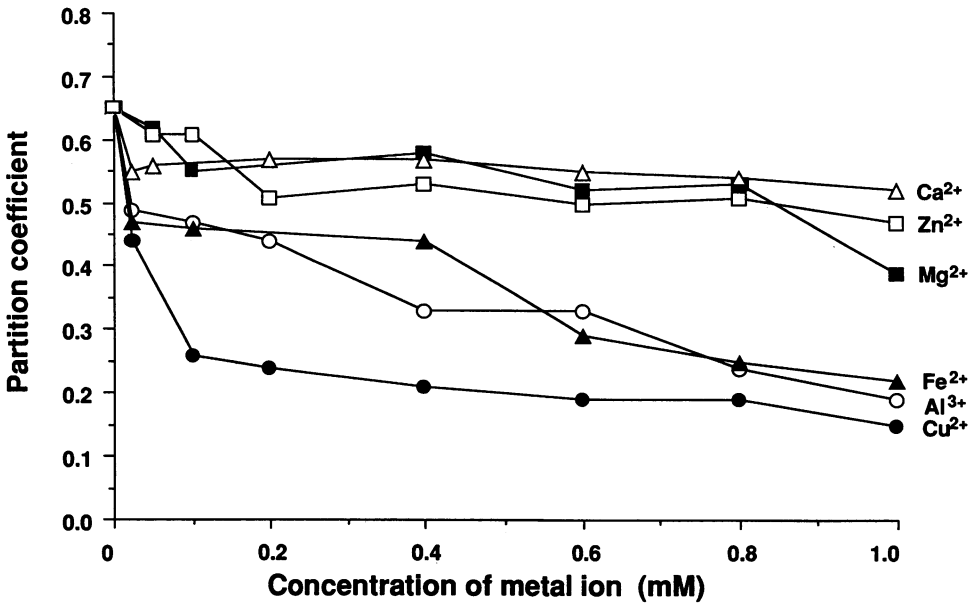

FIG. 1. Changes in partition coefficient of LVFX (0.1 mM) between chloroform and phosphate buffer (pH 5.0) with varying concentrations of $CuSO_4$, $Al(NO_3)_3$, $FeSO_4$, $MgSO_4$, $ZnSO_4$, and $Ca(NO_3)_2$.

control, where partial precipitation of aluminum hydroxide was observed.

The in vitro adsorptive capacities of dried aluminum hydroxide gel for LVFX and OFLX were determined at 37°C. Aluminum hydroxide was suspended in distilled water (2.5 mg/ml), and the pH of the suspension was 7.5. Preliminary experiments showed that the adsorption of LVFX by aluminum hydroxide was rapid since equilibrium was attained within 1 h. An equilibrium time of 1 h was therefore adopted. The results of studies of adsorption of LVFX and OFLX onto aluminum hydroxide are illustrated in Fig. 3, which is plotted according to Langmuir's relationship. The linear regression parameters for the Langmuir treatment are given in Table 2 together with the 95% confidence limits. The adsorptive capacities (r_{max}) and the K values were 7.0 mg/g and 1.77×10^4 M^{-1} for LVFX and 7.4 mg/g and 1.42×10^4 M^{-1} for OFLX, respectively. Aluminum hydroxide adsorbed OFLX and LVFX to almost the same extent.

The rate of adsorption of several quinolones (50 μM) onto aluminum hydroxide in water (2.5 mg/ml) was studied. The percent binding of quinolones followed the order NFLX (72.0% ± 3.29%) > ENX (61.0% ± 3.97%) > OFLX (47.2% ± 1.21%) ≈ LVFX (48.1% ± 3.86%) (mean ± standard deviation, n = 3). NFLX and ENX showed significantly higher binding rates than OFLX and LVFX ($P < 0.05$). Elution of the adsorbed quinolones from aluminum hydroxide was also studied by washing the quinolone-antacid complexes after the adsorption run with distilled water at 37°C for 1 h. The rank order of elution rate of quinolones from aluminum hydroxide was LVFX (17.9% ± 0.57%) ≈ OFLX (20.9% ± 5.58%) ≈ ENX (18.3% ± 2.84%) > NFLX (11.9% ± 1.59%). The elution rate of NFLX was significantly lower than those of other quinolones tested ($P < 0.05$).

TABLE 1. Absorption of LVFX and LVFX-Al^{3+} (1:1) chelate in rats[a]

Sampling time (min)	Amount absorbed (%)	
	LVFX	LVFX-Al^{3+} chelate
10	8.7 ± 1.7	14.0 ± 4.2
20	12.8 ± 0.5	18.3 ± 4.2
30	18.2 ± 5.0	22.6 ± 4.5
45	26.1 ± 6.6	24.0 ± 2.8
60	32.9 ± 6.8	35.2 ± 9.0

[a] Intestinal absorption of drugs (6.75 M) was examined by means of an in situ recirculation experiment; results are expressed as the mean (percent) ± standard deviation (percent) of three animals.

DISCUSSION

There is a large body of information in the literature on the interaction of quinolones with antacids containing certain divalent or trivalent cations. The formation of nonabsorbable chelates has been suggested as the possible mechanism responsible for the decrease in absorption of some quinolones in the presence of these cations (4, 10, 17, 19, 25). However, the magnitude of the decrease in bioavailability varied significantly among the quinolones (13), in spite of their similar stability constants for metal chelate (19). Very little information about mechanisms other than chelate formation for the interaction of quinolones with antacids has been published, and the mechanism remains to be elucidated. This was the background that prompted us to carry out the present study.

The partition coefficient of LVFX was reduced markedly with the addition of Cu^{2+}, Al^{3+}, and Fe^{2+} into aqueous phase (Fig. 1), which indicated the formation of LVFX-metal ion chelate with poor solubility in chloroform layer (17). On the other hand, the binding of LVFX to Mg^{2+}, Zn^{2+}, and Ca^{2+} was much weaker, and these metal ions were shown not to practically form the chelates with LVFX.

The absorption of LVFX and synthetic LVFX-Al^{3+} (1:1) chelate (1) from rat intestine was investigated by the in situ recirculation method. The results indicated that LVFX-Al^{3+}

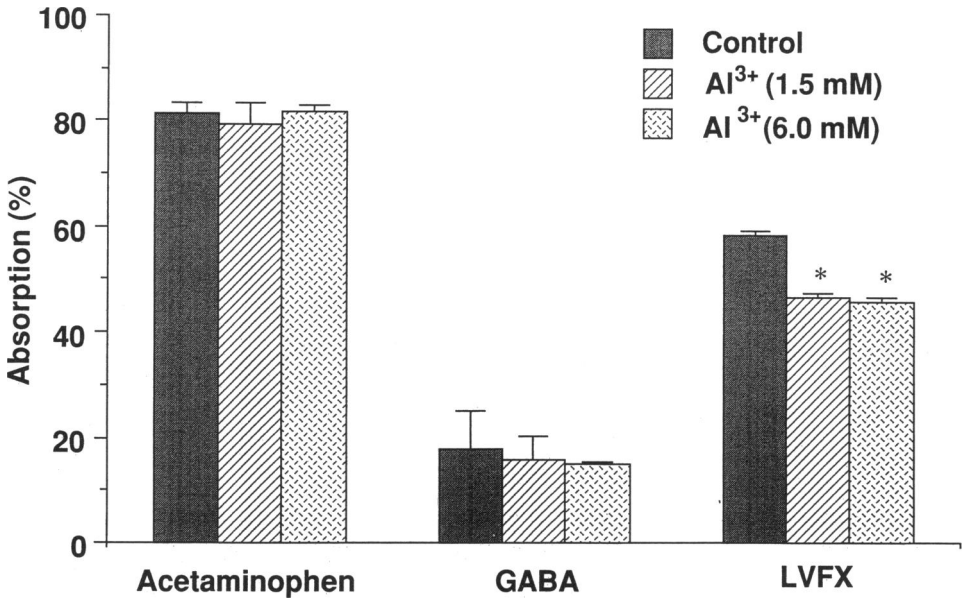

FIG. 2. Effect of $Al(NO_3)_3$ on absorption of acetaminophen, GABA, and LVFX from rat intestinal loop. *, $P < 0.05$, significant different from control.

(1:1) chelate showed no significant difference in absorption from LVFX (Table 1). The phosphate buffer (pH 5.0) was used as aqueous medium in the in vitro partition study and the in situ recirculation experiment, because the stability of LVFX-metal ion chelate complexes is greatest in the weakly acidic pH region (pH 4 to 5) (1). These findings suggested that the stability constant of the LVFX-Al^{3+} (1:1) chelate would not be great enough to reduce significantly absorption of LVFX from rat intestine even at a weakly acidic pH (pH 5.0). It was also suggested that LVFX-Al^{3+} complexes formed in vivo would have different compositions and higher stability compared with the synthetic chelate used in this study.

LVFX is absorbed mainly from the duodenum and small intestine in rats (unpublished data). The pH in the small intestine was reported to be approximately 6.5 (12). Furthermore, it was recently reported that the duodenum and jejunum were the main sites of absorption of ciprofloxacin in humans (26). Thus, the effect of the Al^{3+} ion on absorption of LVFX in rats was investigated further by using the in situ ligated loop method, in which the pH of the drug solution was adjusted to 6.5. It was reported that sparfloxacin was transported in part by a common dipeptide carrier-mediated system in rats (30). We reported previously that LVFX could be absorbed from rat intestine not only by passive diffusion but also by the active carrier-mediated transport system which showed higher affinity to amino acids than to dipeptides (27). The effect of the Al^{3+} ion on these transport systems was studied by using acetaminophen and GABA as model compounds. Acetaminophen, a well-known marker compound for the evaluation of the gastric emptying rate, was reported to be absorbed by passive transport from rat intestine (2). It is thought that GABA would be absorbed by the carrier-mediated system in rats (15). The Al^{3+} ion showed no inhibitory effect on absorption of both compounds, indicating that the passive diffusion and active transport systems in rat intestine were not affected by the Al^{3+} ion. On the other hand, it significantly inhibited absorption of LVFX (Fig. 2). These results seem to suggest that the binding of the Al^{3+} ion to the 4-keto- and 3-carboxyl-groups of quinolones to form nonabsorbable chelates would be responsible for the reduced absorption of quinolones (4, 10, 17, 19, 25).

However, in this experiment, partial precipitation of aluminum hydroxide was observed in the drug solution, when the solution was allowed to stand at room temperature for several minutes. The Al^{3+} ion is particularly insoluble at

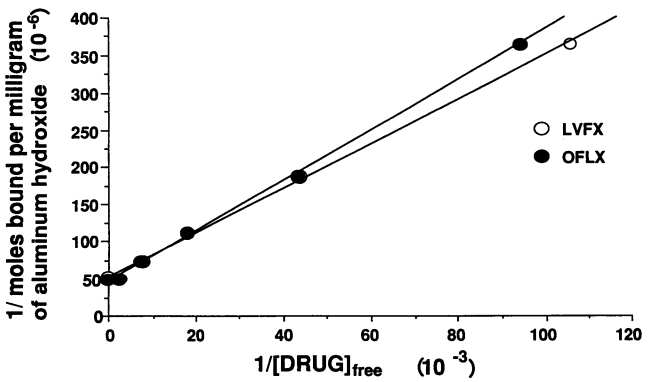

FIG. 3. Langmuir plots: reciprocal of amount of drug bound versus reciprocal of molar free drug concentration for LVFX and OFLX.

TABLE 2. Linear parameters for the Langmuir treatment of the binding of OFLX and LVFX to aluminum hydroxide

Compound	Slope (10^{-3}) (lower/upper limit)[a]	Intercept (10^{-9}) (lower/upper limit)[a]	r_{max} (mg/g)	K (M^{-1})
LVFX	2.98 (2.68/3.28)	0.0527 (0.0369/0.0684)	7.0	1.77×10^4
OFLX	3.44 (3.00/3.88)	0.0489 (0.0282/0.0695)	7.4	1.42×10^4

[a] 95% confidence intervals.

neutral pH as a result of the formation of stable hydroxide complexes, which limit the solubility of Al^{3+} to less than 1 μmol/ml at pH 6.0 (20). The most amorphous forms of aluminum hydroxide are capable of dissolving almost completely in gastric fluid during the normal gastric residence time (9). The soluble aluminum cation, formed in gastric fluid, is reprecipitated in the small intestine when the pH reaches 4.5 or higher. A perfused rat gut experiment recently demonstrated that a large-surface-area amorphous aluminum hydroxide was precipitated in the intestine when an aluminum chloride solution was perfused (20). The published studies (4, 10, 13, 17, 19, 24, 25) do not consider the various forms of aluminum hydroxide which would be present in the small intestine. Antacids including aluminum hydroxide, apart from their neutralizing properties, are known to possess adsorbent properties for endogenous compounds such as bile acids (5, 6, 14) and certain drugs (3, 7, 11). The concomitant ingestion of some drugs with antacids possessing adsorptive properties caused a decrease in bioavailability of the drugs (16). Thus, the in vitro binding properties of LVFX and OFLX for dried aluminum hydroxide gel were studied. LVFX and OFLX were found to be adsorbed by aluminum hydroxide, the adsorption data following a Langmuir plot over the concentration range tested (Fig. 3).

The adsorption of quinolones by aluminum hydroxide followed the rank order NFLX > ENX > OFLX ≈ LVFX. Reversibility of the adsorption process was tested by studying elution of the adsorbed quinolones from aluminum hydroxide. Elution of the adsorbed drugs was not complete (12 to 21%) even after quinolone-antacid complexes were incubated in distilled water at 37°C for 1 h. There is also a difference among the quinolones in their elution rates: LVFX ≈ OFLX ≈ ENX > NFLX. NFLX was more tightly held on aluminum hydroxide than were other quinolones tested. The variation in the extents of adsorption of quinolones may be attributable to the difference in solubility at pH 7.5 of the system. Patrick and Eberman (21) investigated the relation between solubility and adsorption and concluded that, for a given solvent, the more soluble solutes are generally less strongly adsorbed than the less soluble solutes. OFLX and LVFX have much higher solubilities in water than other quinolones (14a). OFLX and LVFX would therefore be expected to be less adsorbed than other quinolones. The results of adsorption and elution experiments indicated that the in vitro interaction of quinolones with aluminum hydroxide followed the rank NFLX > ENX > OFLX ≈ LVFX. The decrease in bioavailability of LVFX and OFLX caused by the simultaneous administration of aluminum hydroxide was much lower than those for ENX and NFLX (23, 24). It seems to be possible to relate the degree of in vitro adsorption of quinolones by aluminum hydroxide to the magnitude of the decrease in bioavailability of various quinolones in vivo. However, this hypothesis should be tested prospectively with other quinolones.

It could be concluded that adsorption of quinolones by aluminum hydroxide reprecipitated in the small intestine would play an important role in the reduced bioavailability of quinolones. The in vitro adsorption studies, particularly when combined with desorption determination, would help in the prediction of the possible interaction that might take place in the gastrointestinal tract following simultaneous or proximal administration of drug combinations. Adsorption of quinolones by various antacids other than aluminum hydroxide is being investigated in our laboratories and will be reported elsewhere in the near future.

REFERENCES

1. **Araki, T., K. Yamazaki, R. Moroi, and H. Hakusui.** 1992. Physicochemical study on the chelate formation of levofloxacin with metal ions, abstr. 663, p. 221. Program Abstr. 32nd Intersci. Conf. Antimicrob. Agents Chemother. American Society for Microbiology, Washington, D.C.
2. **Bagnall, W. E., J. Kelleher, B. E. Walker, and M. S. Losowsky.** 1978. The gastrointestinal absorption of paracetamol in the rat. J. Pharm. Pharmacol. **31**:157–160.
3. **Blaug, S. M., and M. R. Gross.** 1965. In vitro adsorption of some anticholinergic drugs by various antacids. J. Pharm. Sci. **54**: 289–294.
4. **Campbell, N. R. C., M. Kara, B. B. Hasinoff, W. M. Haddara, and D. W. McKay.** 1992. Norfloxacin interaction with antacids and minerals. Br. J. Clin. Pharmacol. **33**:115–116.
5. **Clain, J. E., J. Malagelada, V. S. Chadwick, and A. F. Hofmann.** 1977. Binding properties in vitro of antacids for conjugated bile acids. Gastroenterology **73**:556–559.
6. **Eaton, L., J. L. White, and S. L. Hem.** 1991. In-vitro adsorption of cholate anion at pH 7.5 by aluminum hydroxide. Drug Dev. Ind. Pharm. **17**:2029–2039.
7. **El-Masry, S., and S. A. H. Khalil.** 1974. Adsorption of atropine and hyoscine on magnesium trisilicate. J. Pharm. Pharmacol. **26**:243–248.
8. **Hayakawa, I., S. Atarashi, S. Yokohama, M. Imamura, K. Sakano, and M. Furukawa.** 1986. Synthesis and antibacterial activities of optically active ofloxacin. Antimicrob. Agents Chemother. **29**:163–164.
9. **Hem, S. L., J. L. White, J. D. Buehler, J. R. Luber, W. M. Grim, and E. A. Lipka.** 1982. Evaluation of antacid suspensions containing aluminum hydroxide and magnesium hydroxide. Am. J. Hosp. Pharm. **39**:1925–1930.
10. **Issopoulos, P. B.** 1989. Spectrophotometric determination of trace amounts of iron(III) with norfloxacin as complexing reagent. Analyst **114**:627–630.
11. **Khalil, S. A., N. A. Daabis, V. F. Naggar, and M. M. Motawi.** 1976. The in vitro adsorption of some antibiotics on antacids. Pharmazie **31**:105–109.
12. **Koizumi, T., T. Arita, and K. Kakemi.** 1964. Absorption and excretion of drugs. XX. Some pharmacokinetic aspects of absorption and excretion of sulfonamides (2). Absorption from rat small intestine. Chem. Pharm. Bull. **12**:421–427.
13. **Lomaestro, B. M., and G. R. Bailie.** 1991. Quinolone-cation interactions: a review. DICP Ann. Pharmacother. **25**:1249–1258.
14. **Mangnall, Y. F., A. Smythe, and A. G. Johnson.** 1986. The ability of antacids and cholestyramine to bind bile acids: effect of pH. Scand. J. Gastroenterol. **21**:789–794.

14a. **Matsumoto, J.** 1991. Physicochemical properties and pharmacokinetics of quinolones, p. 67–76. *In* S. Mitsuhashi (ed.), New quinolones. Japan Scientific Society Press, Tokyo.

15. **Merino, M., A. Polache, J. E. Peris-Ribera, J. Moreno-Dalmau, and J. M. Pla-Delfina.** 1989. Absorption kinetics of β-alanine as model compound in rat small intestine. Biochim. Biophys. Acta. **986**:1–7.
16. **Moustafa, M. A., H. I. Al-Shora, M. Gaber, and M. W. Gouda.** 1987. Decreased bioavailability of quinidine sulphate due to interaction with adsorbent antacids and antidiarrhoeal mixtures. Int. J. Pharm. **34**:207–211.
17. **Nakano, M., M. Yamamoto, and T. Arita.** 1978. Interactions of aluminum, magnesium, and calcium ions with nalidixic acid. Chem. Pharm. Bull. **26**:1505–1510.
18. **Okazaki, O., H. Aoki, and H. Hakusui.** 1991. High-performance liquid chromatographic determination of (S)-(−)-ofloxacin and its metabolites in serum and urine using solid-phase clean-up. J. Chromatogr. **563**:313–322.
19. **Okazaki, O., T. Kurata, and H. Tachizawa.** 1988. Studies on the mechanism of pharmacokinetic interaction of aluminum hydroxide, an antacid, with new quinolones in rats. Xenobio. Metab. Dispos. **3**:387–394.
20. **Partridge, N. A., F. E. Regnier, J. L. White, and S. L. Hem.** 1989. Influence of dietary constituents on intestinal absorption of aluminum. Kidney Int. **35**:1413–1417.

21. **Patrick, W. A., and N. F. Eberman.** 1925. Studies in adsorption from the standpoint of capillarity—II. J. Phys. Chem. **29**:220–228.
22. **Sato, K., Y. Matsuura, M. Inoue, T. Une, Y. Osada, H. Ogawa, and S. Mitsuhashi.** 1982. In vitro and in vivo activity of DL-8280, a new oxazine derivative. Antimicrob. Agents Chemother. **22**:548–553.
23. **Shiba, K., A. Saito, T. Miyahara, H. Tachizawa, and T. Fujimoto.** 1988. Effect of aluminum hydroxide, an antacid, on the pharmacokinetics of new quinolones in humans. Xenobio. Metab. Dispos. **3**:717–722.
24. **Shiba, K., O. Sakai, J. Shimada, O. Okazaki, H. Aoki, and H. Hakusui.** 1992. Effect of antacids, ferrous sulfate, and ranitidine on absorption of DR-3355 in humans. Antimicrob. Agents Chemother. **36**:2270–2274.
25. **Shimada, J., K. Shiba, T. Oguma, H. Miwa, Y. Yoshimura, T. Nishikawa, Y. Okabayashi, T. Kitagawa, and S. Yamamoto.** 1992. Effect of antacid on absorption of the quinolone lomefloxacin. Antimicrob. Agents Chemother. **36**:1219–1224.
26. **Staib, A. H., D. Beerman, S. Harder, U. Fuhr, and D. Liermann.** 1989. Absorption differences of ciprofloxacin along the human gastrointestinal tract determined using a remote-control drug delivery device (HF-capsule). Am. J. Med. **87**(Suppl. 5A):66–69.
27. **Tanaka, M., T. Kurata, Y. Ohshima, H. Aoki, O. Okazaki, and H. Hakusui.** 1992. The mechanistic study on inhibition of levofloxacin (LVFX) absorption by metal ions, abstr. 662, p. 221. Program Abstr. 32nd Intersci. Conf. Antimicrob. Agents Chemother. American Society for Microbiology, Washington, D.C.
28. **Tsuji, A., E. Miyamoto, and E. Kagami.** 1978. GI absorption of β-lactam antibiotics I: kinetic assessment of competing absorption and degradation in GI tract. J. Pharm. Sci. **67**:1701–1704.
29. **Une, T., T. Fujimoto, K. Sato, and Y. Osada.** 1988. In vitro activity of DR-3355, an optically active ofloxacin. Antimicrob. Agents Chemother. **32**:1336–1340.
30. **Yamaguchi, T., M. Yokogawa, Y. Sekine, and M. Hashimoto.** 1991. Intestinal absorption characteristics of sparfloxacin. Xenobio. Metab. Dispos. **6**:53–59.